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DESIGN, SYNTHESIS AND EVALUATION OF CATECHOL-BASED AMYLOID BETA AGGREGATION INHIBITORS

Hiroyuki Konno[a]* and Kenta Teruya[b]

[a]Department of Chemistry and Biological Engineering, Graduate School of Science and Engineering, Yamagata University, Yonezawa, Yamagata 992-8510, Japan. [b]Graduate School of Medicine, Tohoku University, Aoba-ku, Sendai 981-8575, Japan. *E-mail address: konno@yz.yamagata-u.ac.jp

Abstract – The use of small molecules to inhibit amyloid β aggregation is one approach under investigation to prevent the onset of Alzheimer's disease (AD). One such small molecule is curcumin, but its utility is limited by poor water solubility and hence poor bioavailability. In this review, we summarize the findings of structure-activity relationship studies of curcumin and diaryl γ-dihydropyrone, undertaken to improve their inhibitory effect and physical properties. The key findings are: phenolic hydroxy groups can be introduced at the 2 and 3,4-positions of phenol rings, the C7 spacer of curcumin is essential for good anti-amyloid β aggregation activity; and the catechol moiety is important for both anti-aggregation activity and water solubility. The combination of the C5 spacer unit and mono ketone of curcumin analogues is more effective for water solubility with high anti-amyloid β aggregation activity. Diaryl γ-dihydropyrone with hydroxy groups on phenol rings as cyclocurcumin mimetics also showed anti-amyloid β aggregation activity and high water solubility. Analogues of berberine, a benzylisoquinoline alkaloid also of interest to treat central nervous system diseases, are also reviewed. Finally, we review efforts to develop fluorescent curcumin analogues for use as diagnostic agents.

CONTENTS

1. INTRODUCTION

The curcumins[1] are components of turmeric, which is widely used in curry and in traditional Indian medicines to treat biliary disorders, anorexia, and coughs. The most common of the curcumins is curcumin 1 (CUR, **1**) [(1*E*,6*E*)-1,7-bis(4-hydroxy-3-methoxyphenyl)hepta-1,6-diene-3,5-dione], usually referred to as curcumin, but curcumin 2 (**2**) [desmethoxycurcumin] and curcumin 3 (**3**) [bis-desmethoxycurcumin] are also known, all of which form stable enols and are responsible for the yellow color of turmeric. Multiple studies have established CUR (**1**) as an antioxidant and anti-inflammatory, and it has been hypothesized to have anti-Alzheimer's disease activity (Figure 1).[2-4]

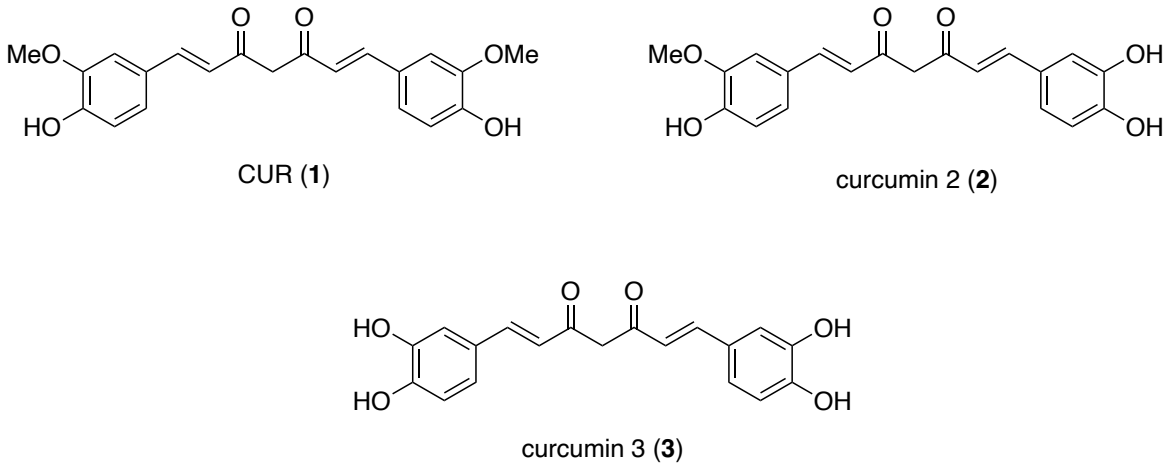


Figure 1. Chemical structures of CUR (**1**), curcumin 2 (**2**), and curcumin 3 (**3**)

Previous studies identified a toxic peptide,[5] amyloid beta (Aβ) , as the cause of AD pathogenesis.[6,7] Aβ is generated by the metabolism of amyloid precursor protein, which is cleaved by the both β and γ-secretases from within the brain and is prone to form toxic aggregates.[8] Drugs capable of dissociating Aβ aggregates therefore hold promise as to treat AD.[9,10] One such drug, Aducanumab (aduhelm) was FDA approved in 2021 based on its ability to decrease levels of Aβ aggregates in patients with MCI (mild cognitive

impairment).[11,12] However, its uncertain effectiveness and relatively high cost have proved controversial (Figures 2 and 3).

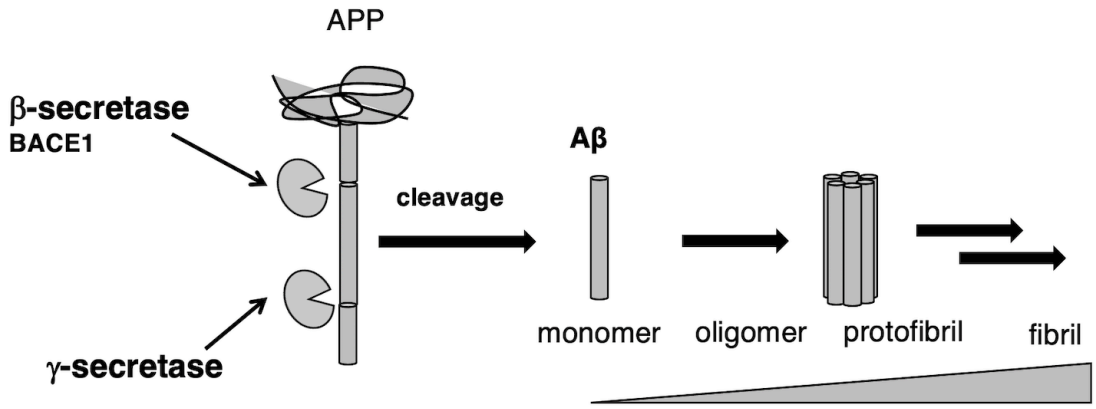


Figure 2. The formation of amyloid fibrils by the synthesis and aggregation of Aβ42

Figure 3. Sequence of Aβ42

Other drugs used to treat AD such as acetylcholine esterase (AChE) inhibitors and *N*-methyl-D-aspartic acid (NMDA) receptor antagonists are not disease-modifying and address only its symptoms.[13] Examples of these include donepezil (**4**),[14] memantine (**5**),[15] galanthamine (**6**)[16] and rivastigmine (**7**)[17] (Figure 4).

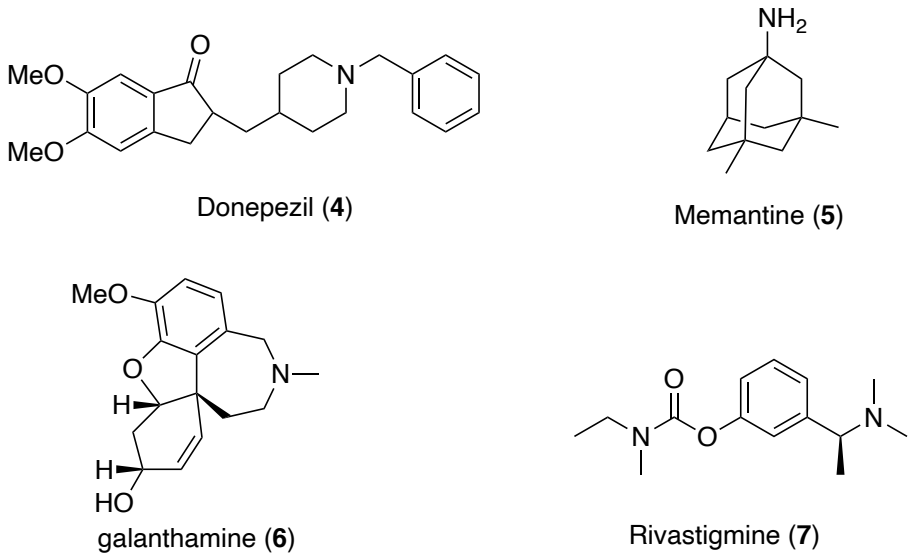


Figure 4. Chemical structures of donepezil (**4**), memantine (**5**), galanthamine (**6**) and rivastigmine (**7**)

CUR (**1**) inhibited Aβ formation and lowered Aβ levels in the brains of mice after intraperitoneal administration,[18] but the failure to have a similar effect in AD patients, perhaps because of its poor water solubility. A great deal of effort has therefore been invested in the development of water-soluble derivatives of CUR (**1**). Early studies resulted in CUR 4β-D-glucuronide (**8**),[19] CUR sulphate, and a selection of reduced CUR derivatives, but these were rapidly metabolized by the liver, disqualifying them from further development.[20] Subsequent work resulted in the synthesis of hybrid compounds comprising CUR (**1**) conjugated with another moiety such as glucose. For example, Dolai et al. reported the CUR-type compound (**9**) bearing a PEG-linked glucose;[21] and Zhang reported that the cholesterol-bearing CUR hybrid (**10**) demonstrated an inhibitory effect on Aβ aggregation (Figure 5).[22,23] Compound **10** was also found to localize to the cell membrane/lipid rafts (CM/LR) of human blastoma MC65 cells, protecting them from Aβ fibrils, and was to have the potential to cross the blood-brain barrier (BBB). Ouberai et al. reported CUR dimer (**11**) comprising CUR (**1**) bound to a cyclopeptide scaffold, and that it could inhibit Aβ fibril formation.[24] Despite these advances, the significant increase in the molecular weight of the hybrid compounds compared with CUR (**1**) is considered a disadvantage (Figure 5).

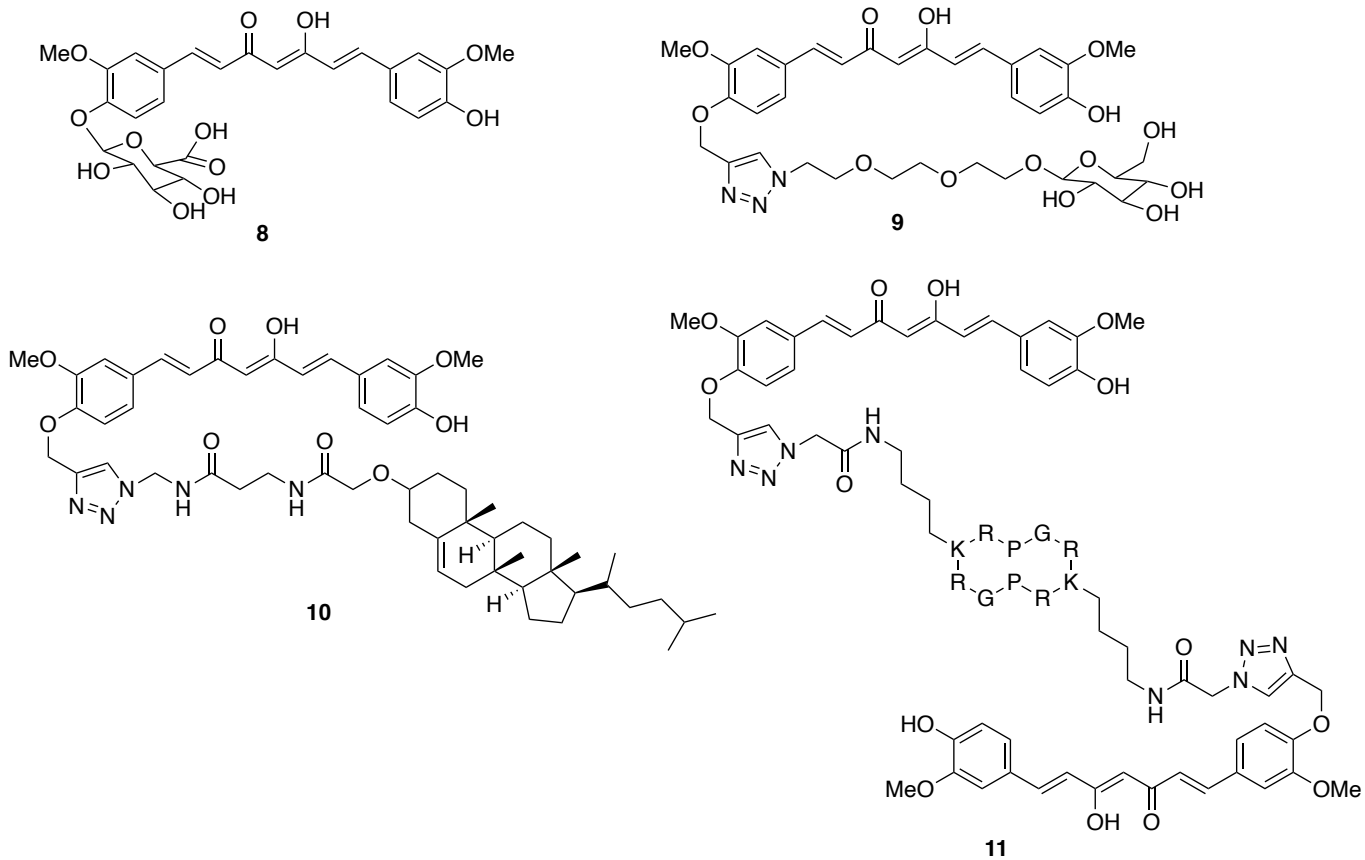
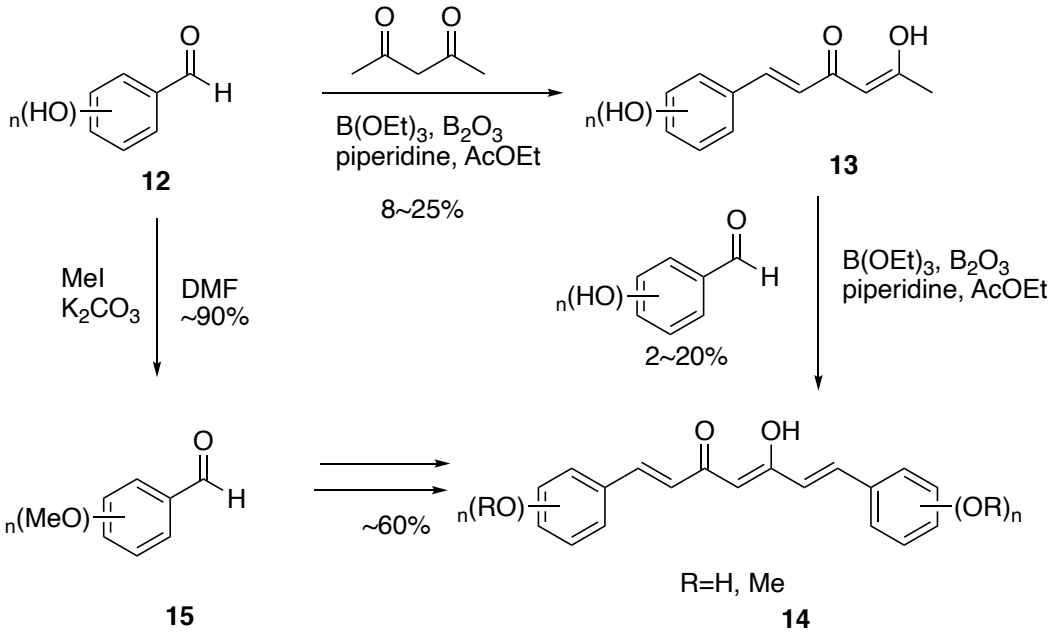


Figure 5. Chemical structures of CUR 4β-D-glucuronide (**8**), Dolai's compound (**9**), steroidal compound (**10**), and CUR dimer (**11**)

In this review paper, we report the optimization of CUR analogues and their related compounds for their ability to inhibit Aβ aggregation. A prerequisite for this project is an understanding of the structural properties of CUR (**1**), and the identification of the specific pharmacophore of the CUR (**1**) framework responsible for the inhibition. The scope of our study extended to C5-curcumin analogues and cyclocurcumin pyran type analogues, as well as analogues of the alkaloid berberine, which is highly water soluble. The use of CUR analogues as fluorescence probes to aid in the diagnosis of AD is also disclosed.

2. CURCUMINS: SYNTHESIS AND ANTI-AMYLOID BETA AGGREGATION ASSAY

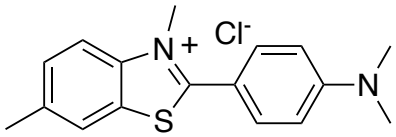
The curcumins incorporate olefin, β-diketone moieties, and phenol rings moieties, and adopt planar structures owing to extensive π-conjugation between the olefin and β-diketone.[1,25] They can be synthesized via the aldol reaction of an appropriate benzaldehyde and diketone using Pabon's protocol,[26] as conventional aldol reaction conditions to afford mainly 3-substituted-2,4-diketone derivatives. Treatment of benzaldehyde derivatives (**12**) and 2,4-pentanedione with $(EtO)_3B/B_2O_3$/piperidine in dioxane gave **13** in 8-25% yields, with 3-benzyl-2,4-pentanedione as a by-product, which was removed by chromatography. Condensation of the second benzaldehyde, also using Pabon's protocol, affords the desired CUR derivatives (**14**) in 2-20% yields. The aldol condensations proceed in higher yields if protected benzaldehydes are used, but the difficulties associated with the deprotection of phenolic groups precludes the use of this strategy (Scheme 1).[27]



Scheme 1. General method for the preparation of curcumin analogues **14**

The extent of Aβ aggregation inhibition demonstrated by these derivatives was assessed using the Thioflavin T (**16**, ThT) method, which is based on the fluorescence of ThT (**16**) when bound to Aβ

aggregation. The intensity of the fluorescence is proportional to the concentration of fibrils present, enabling the inhibitory effect of an added compound to be estimated.[28-31] When incubated with 25 μM of freshly-prepared amyloidogenic Aβ42 at 37 ºC, and after an initiation lag of 2 h, the fluorescence of ThT (**16**) increased, following a sigmoidal-like curve with a point of inflection and reaching a maximum after 4 h to 6 h. Although the final equilibrium level was achieved within this time, the extent of Aβ42 aggregation after 20 h was defined to be 100%. When the mixture of 25 μM Aβ42 and 10 μM CUR (**1**) was incubated at 37 ºC, the fluorescence followed a similar sigmoidal-like curve, with a final equilibrium level of ~20%. Therefore, the inhibition of Aβ aggregation by CUR (**1**) was calculated to be 80%.[32] Treatment of Aβ42 with CUR (**1**) at 20 h was defined as about ~20% aggregate as a control and accordingly following assay was evaluated (Figure 6).



Thioflavin T (**16**)

Figure 6. Chemical structure of thioflavin T (**16**)

3. CURCUMIN ANALOGUES I

Having used the ThT (**16**) method to assess the inhibition of Aβ aggregation by CUR (**1**), we undertook a structure-activity relationship study of water-soluble CUR (**1**) analogues (Figure 7). First, we sought to determine whether the bis-phenols could be substituted for free hydroxy groups, and the importance of the ketone and double-bond moieties. Structure-activity relationship studies established that analogues incorporating a catechol-containing pharmacophore inhibited Aβ aggregation, and the molecular modifications that tolerated by CUR analogues with such a catechol motif. Our design of these analogues is expected to result in the availability of bio-probes to investigate Alzheimer's disease.

We also discovered two new approaches to improving the water solubility of CUR analogues: (a) reducing their molecular planarity, and (b) adding a β-diketone moiety. The *o*-phenolic hydroxy group present in the CUR analogues was found to play the important role of stabilizing the *o*-phenol ring and neighboring olefin. This work resulted in inhibitor (**21**), which demonstrated improved low water solubility compared with CUR (**1**), and better inhibitory activity. The knowledge gathered in this study should inform the development of new approaches to improving the water solubility of curcumin-type compounds, whose in vivo use is expected (Figure 7).[33]

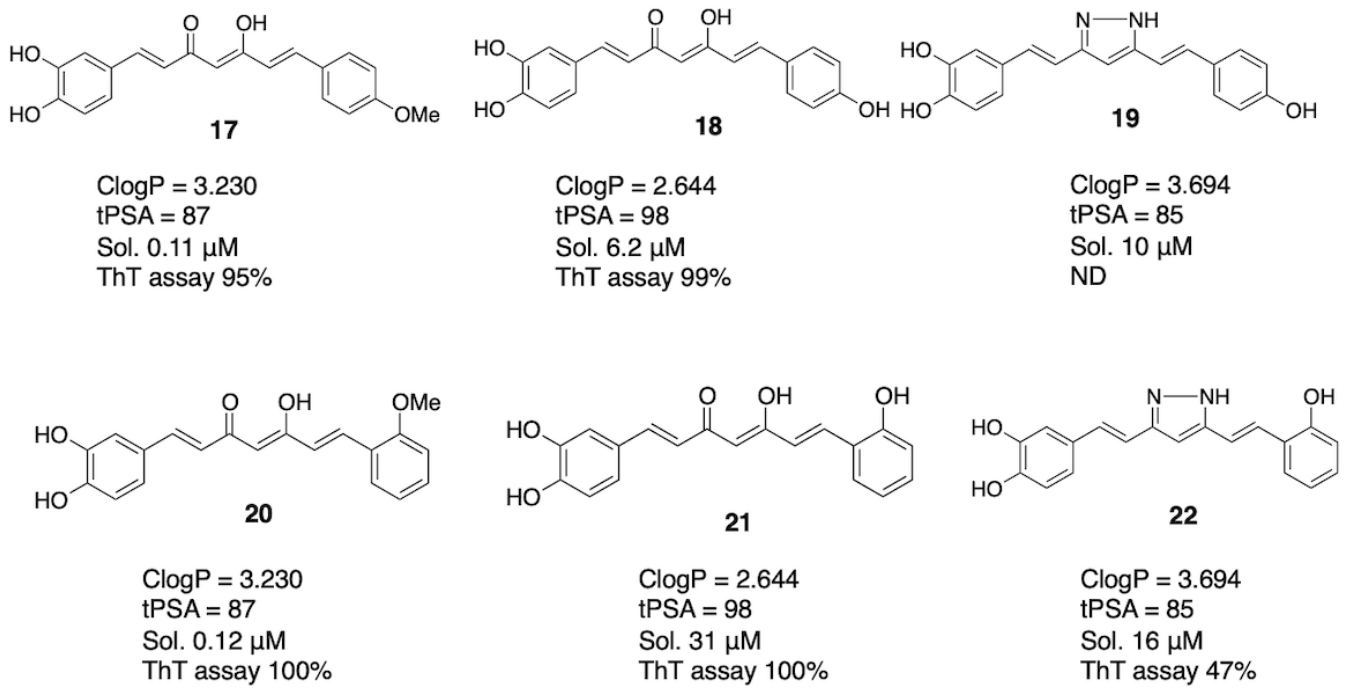


Figure 7. Structure-activity relationship study of water-soluble CUR analogues (**17**-**22**)
ClogP: c-logarithm partition coefficient, tPSA: topological Polar Surface Area, Sol.: water solubility.

We also discovered that the CUR framework is a non-peptidyl inhibitor of beta-site amyloid precursor protein cleaving enzyme 1 (BACE1), although the IC_{50} values of candidate CUR derivatives were moderate (~250 μM).[34] Another structure-activity relationship study of CUR derivatives[32,35] led to the conclusion that phenolic hydroxy groups and an alkenyl spacer are important structural features[36,37] needed for BACE1 inhibition, and the inhibition of enzyme activity by these derivatives is non-competitive based on the results of a kinetics experiment and docking simulation. Two polar phenolic hydroxy groups and a ketone of **21** and **23** were involved in hydrogen bonding interactions (Gly230 and Glu339) by docking simulations using MOE software. Especially, *o*-phenolic hydroxy group and indolyl amino group were important functionalities (Figure 8).[38]

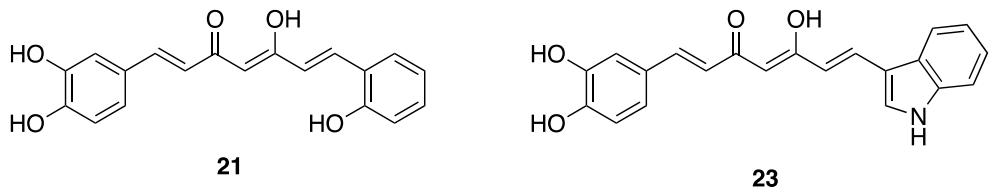


Figure 8. Chemical structures of **21** and **23** against BACE1

The Murray group reported that aminopyridine derivatives (**24**,**25**) inhibit the active site of aspartyl protease;[39,40] the aminopyridine motif upon which they are based was found by combination of fragment screening techniques and high throughput X-ray crystallography against BACE1.[41-42] We designed BACE1

inhibitors (**26**) and (**27**) incorporating the C5 spacer connected with the amino pyridine moiety and 3-methoxyphenyl groups for potent inhibitory activities as CUR mimetics (Figure 9).

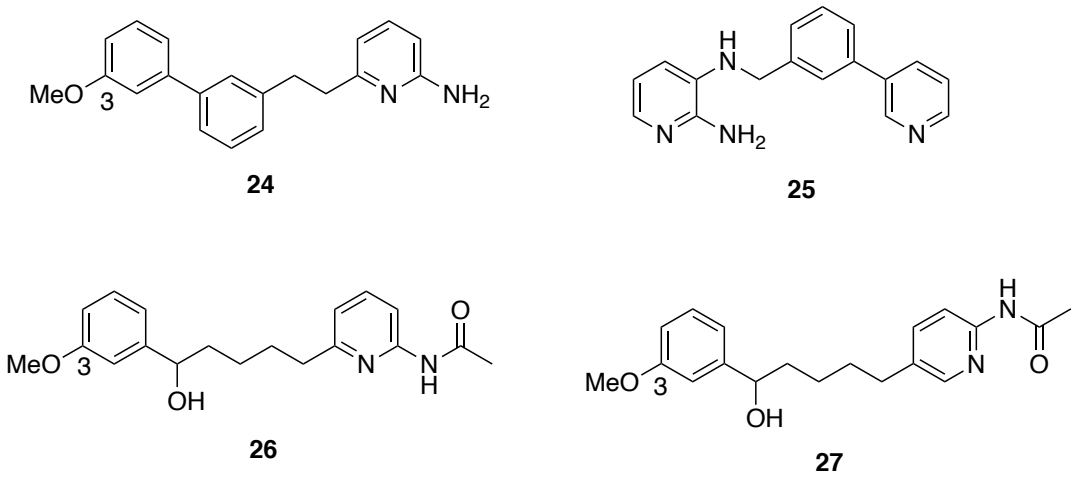
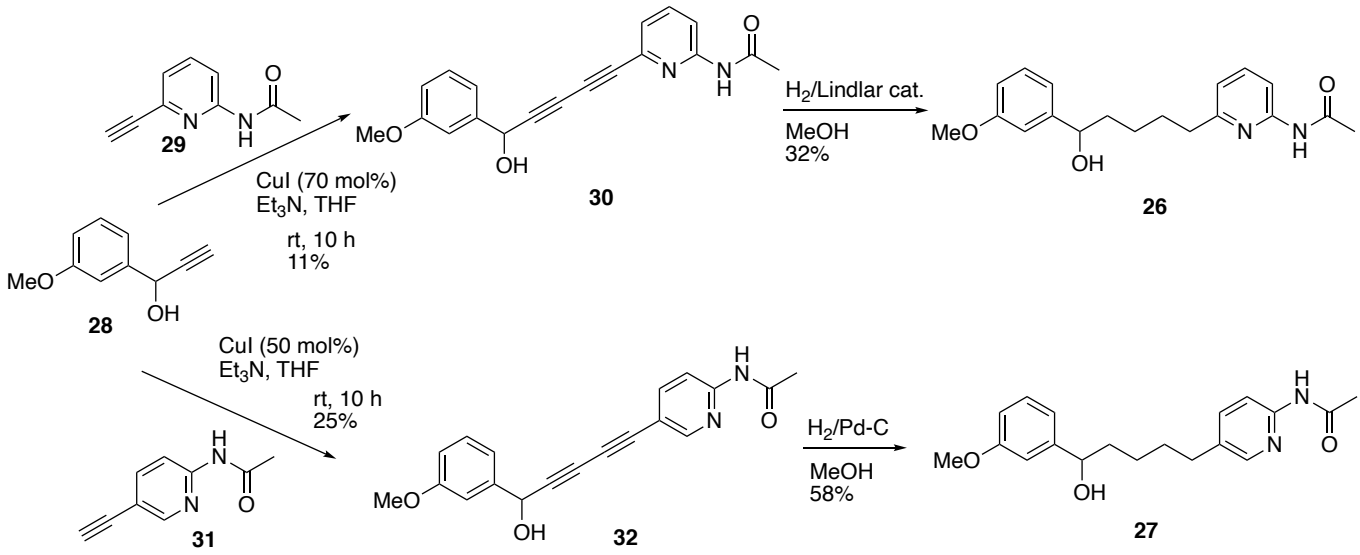


Figure 9. BACE1 inhibitors (**24-27**) with an aminopyridine moiety

Compounds **26** and **27** were prepared by the Glaser homo coupling reaction of terminal alkynes.[43,44] Treatment of **28** and either **29** or **31** in the presence of CuI in THF gave diyne (**30**) or (**32**) in low yields, which were hydrogenated to give the target compounds (**26)** and (**27)** in moderate yields; the low yields of **30** and **32** can be attributed to their poor solubility in organic solvents. 6-Alkyl-2-aminopyridine (**26**) showed greater inhibitory activity against BACE1 in comparison with 5-alkyl-2-aminopyridine (**27**). Aminopyridine derivatives, which had an acetyl group removed of **26**, showed similar activity (Scheme 2).[45]



Scheme 2. Synthesis of aminopyridine type BACE1 inhibitors (**26**) and (**27**)

4. CURCUMIN ANALOGUES II

In section 3, we described how the anti Aβ aggregation properties of CUR (**1**) can be improved by substituting free hydroxy groups of the bis-phenols; and that both the ketone and double bond moieties in the spacer are essential for inhibitory activity. Based on these findings, we synthesized the 2',3,4-trihydroxy derivative (**21**), a highly water-soluble inhibitor of Aβ aggregation.[33]

We sought to investigate the importance of the length and the number of carbonyl groups contained in the spacer unit. A variety of CUR derivatives were synthesized, including the C7-spacer-containing 2,3-dihydroxy derivative (**33**) and the 3,4-dihydroxy derivative (**34**). The water solubility of 3,4-dihydroxy derivative (**34**) was 20 times higher than that of 2,3-dihydroxy derivative (**33**) despite having similar toxicities and behaving similarly in the ThT assay. Compound **35** could not be isolated due to its susceptibility to undergo a retro-aldol reaction. However, compound (**37**) demonstrated excellent Aβ aggregation inhibition and water solubility. Yields of the chalcone derivatives (**38**) and (**39**) bearing with mono-ketone C3-spacers were moderate (Figure 10).

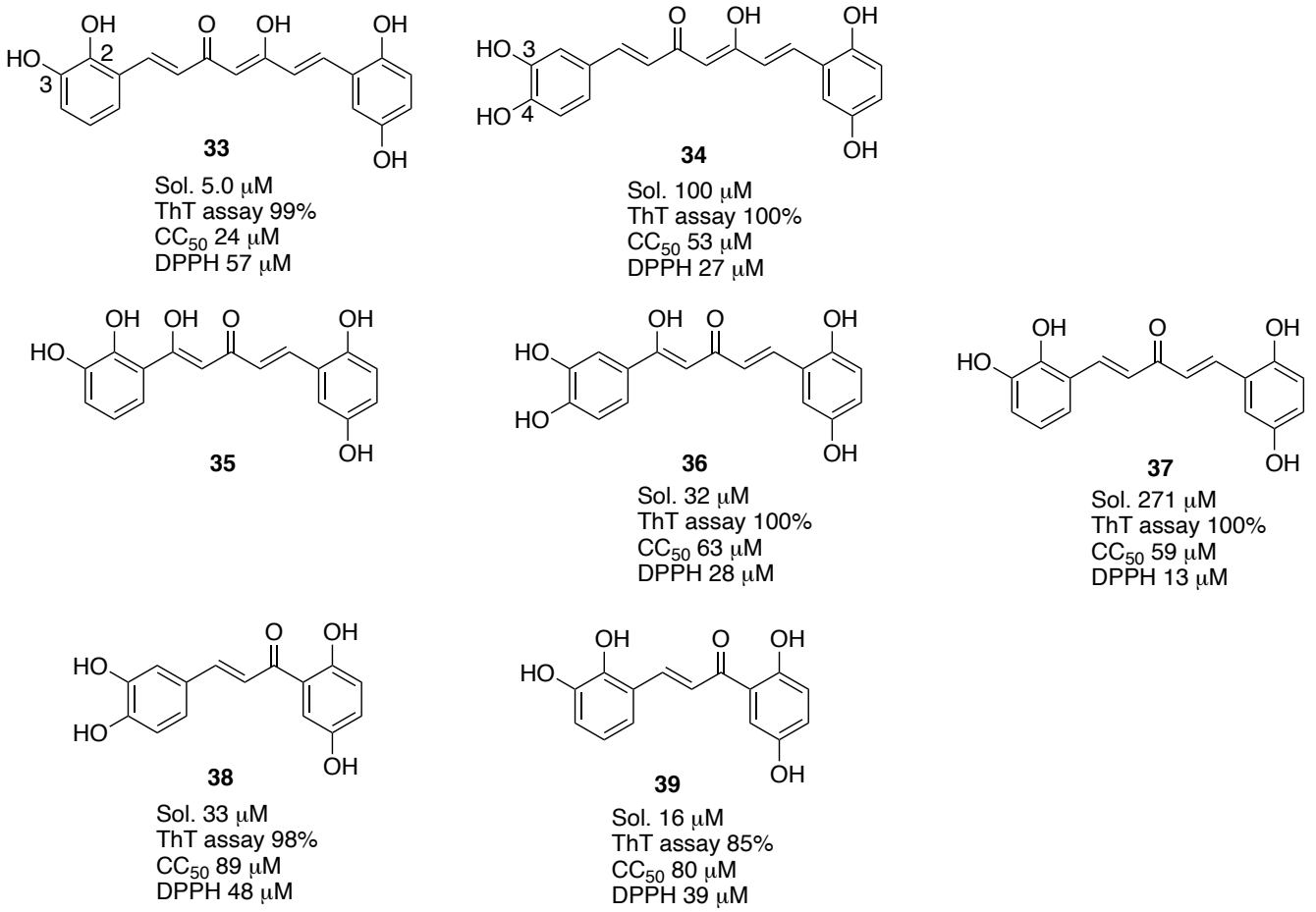
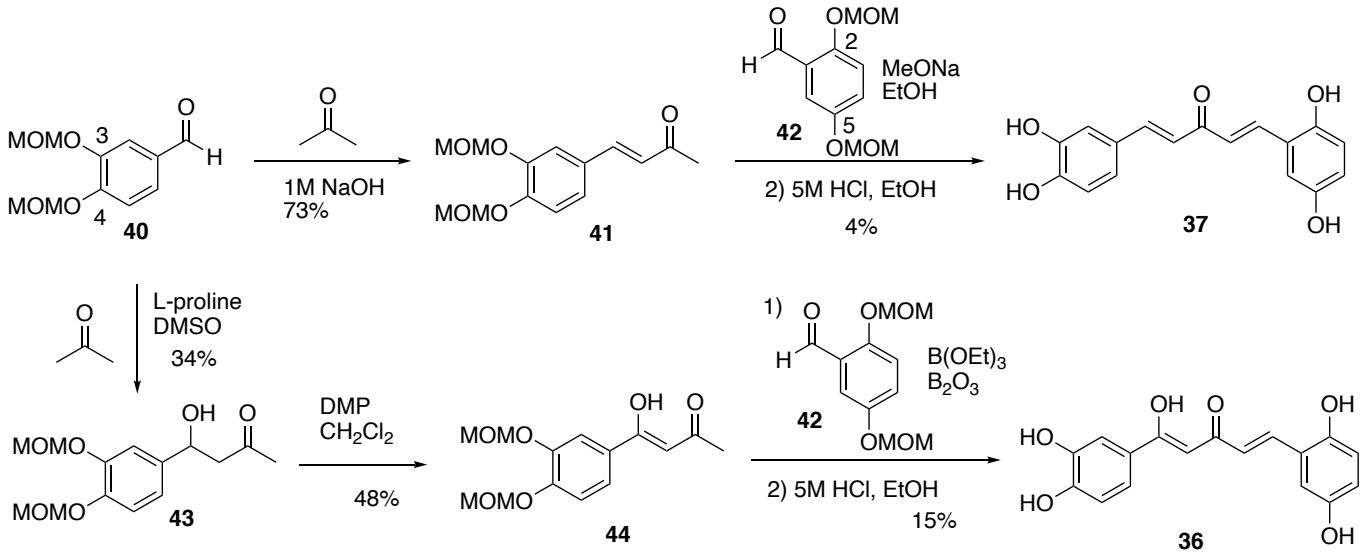


Figure 10. Molecular design adopted for water-soluble Aβ aggregation inhibitors

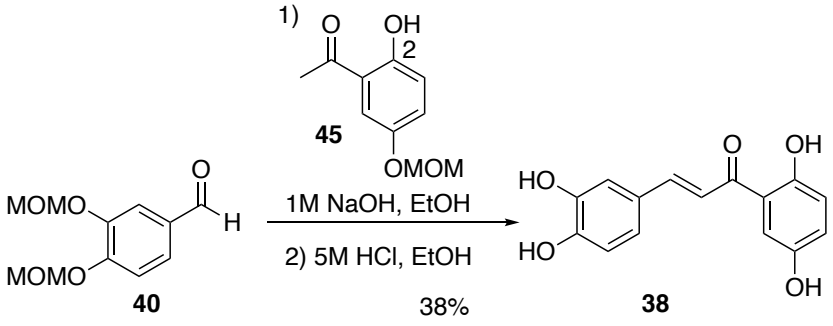
ClogP: c-logarithm partition coefficient, Sol.: water solubility, CC_{50}: cytotoxicity concentration of 50%, DPPH: 2,2-diphenyl-1-picrylhydrazyl radical scavenger activity assay.

The C5- and C3-spacer type CUR derivatives (**36**), (**37**), and (**38**) were also prepared (Schemes 3 and 4). Treatment of acetone and 3,4-(dimethoxymethoxy)benzaldehyde (**40**) in the presence of NaOH gave unsaturated ketone (**41**) in yields of 73%. In contrast, the proline-catalyzed aldol reaction[46] of acetone and **40** afforded **43** in a yield of 34%. Aldol reaction between the unsaturated ketone (**41**) and 2,5-(dimethoxymethoxy)benzaldehyde (**42**) with MeONa followed by the deprotection of MOM ethers gave the corresponding C5-monoketone CUR analogue (**37**) in 4% yield. Dess-Martin periodinane oxidation of β-hydroxyketone **43** gave diketone **44** in 48% yield, which was transformed C5-diketone CUR analogue (**36**) in 15% yield. The very low yields of these reactions were attributed to the steric hindrance of the MOM ether moiety on the 2-position of **42** impeding access to the aldehyde group (Scheme 3).[47]



Scheme 3. Synthesis of C5-CUR analogues (**36**) and (**37**)

Aldol condensation of **40** and **45** followed by the deprotection of MOM groups yielded chalcone derivative (**38**) in 38% yield. However, by using **45** as a coupling partner, chemical yield of the aldol reaction was improved (Scheme 4).



Scheme 4. Synthesis of chalcone derivative (**38**)

Five compounds including CUR (**1**), Me-CUR (**71**), C5-CUR (**37**), and chalcone (**38**) and (**39**), exhibited inhibition of abnormal prion protein propagation in persistently prion-infected cells.[48] Their structure-activity relationships indicate that their hydrogen bonding and hydrophobicities contribute to their biological activities much more than their size.

5. DIARYL γ-DIHYDROPYRONE TYPE COMPOUNDS

In previous work, we found that the C5-CUR (**37**) comprising two phenolic rings connected by C5-sp^2 carbons exhibited high Aβ aggregation inhibitory activity and adequate water solubility. However, CURs incorporating sp^2 carbons are notoriously unstable in water. Cyclocurcumin (CYC, **46**) reported by Kiuchi et al. is a minor CURs formed by intramolecular Michael addition of CUR (**1**) and shown to have a synergistic effect with CUR (**1**) as a nematicide and inhibitor of the growth of human breast cancer MCF-7 cells.[49-51] Our pursuit of water soluble diaryl γ-dihydropyrone type derivative (**47**) with anti Aβ aggregation activity and stability by mimicking CYC (**46**) was reported (Figure 11).

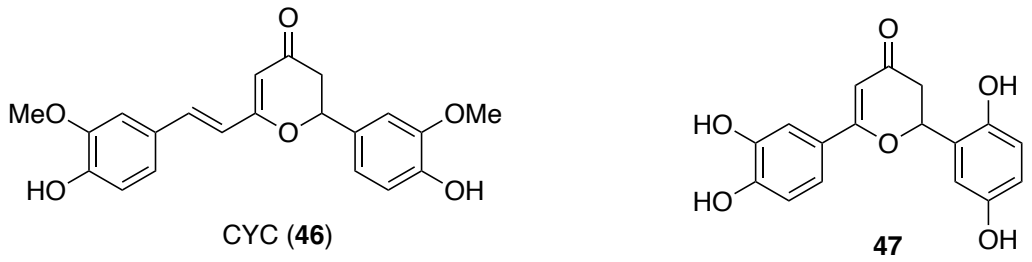
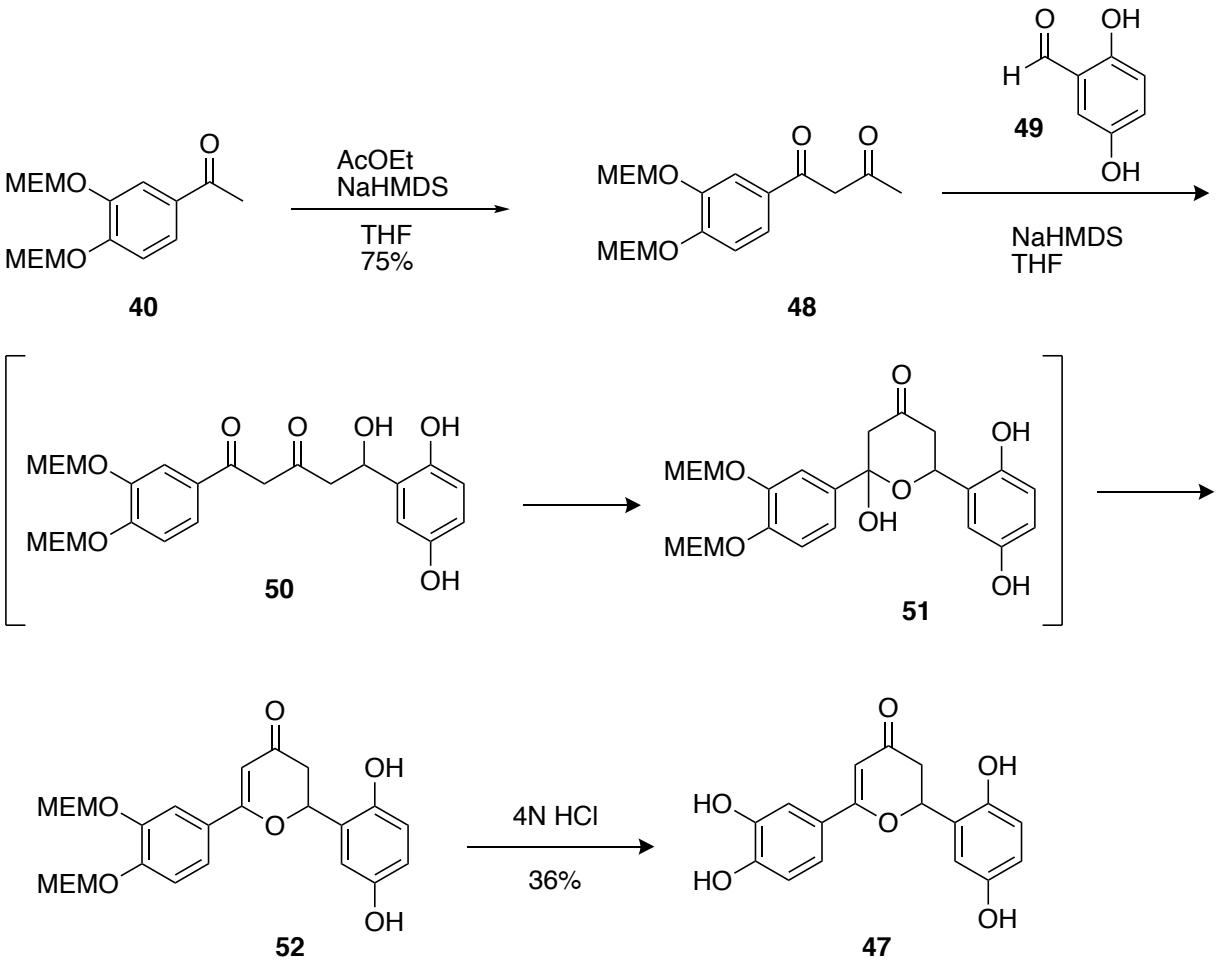


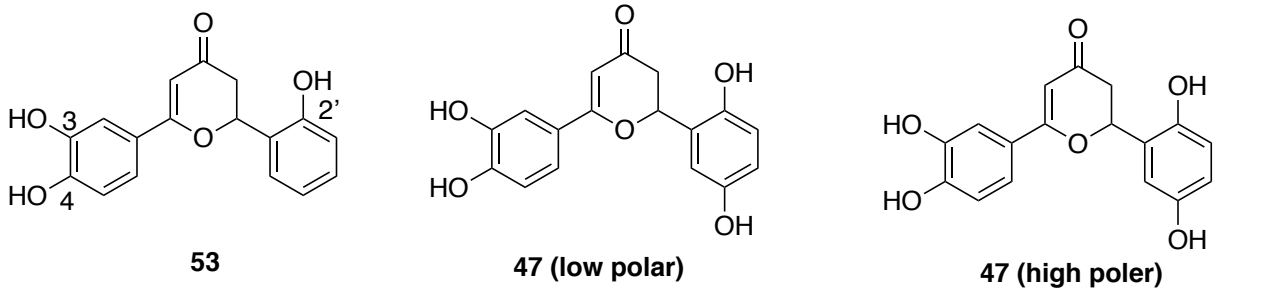
Figure 11. Chemical structure of CYC (**46**) and diaryl γ-dihydropyrone (**47**)

A series of γ-dihydropyrone derivatives (**47**) was synthesized according to Khera's method.[52] Aldol reaction of acetophenone (**40**) with ethyl acetate in the presence of NaHMDS afforded the diketone derivative (**48**), which was treated with NaHMDS and then a variety of benzaldehyde (**49**) to give the corresponding hydroxy diketone (**50**) and then dehydration via γ-tetrahydropyrone (**51**) to give γ-dihydropyrone (**52**). After HCl-promoted cyclization and dehydration, the requisite aryl γ-dihydropyrone (**52**) was obtained. Deprotection of the MEM groups using 4M HCl yielded the desired diaryl γ-dihydropyrone (**47**) (Scheme 5).

Hybrid compound **53** incorporates two phenolic rings bearing hydroxy groups at the 3,4,2'-positions and showed moderate Aβ aggregation inhibition with good water solubility (259 μM). Diaryl γ-dihydropyrones (**47**) were also potent Aβ aggregation inhibitors at 1 μM, and showed good water solubility (2339 μM) and low cytotoxicity. The constituent enantiomers of **47** behaved similarly to the racemate. Both compounds were identified to the comparison of stable conformers by molecular mechanism experiments (Figure 12).[53]



Scheme 5. Synthesis of γ-dihydropyrone type compound (**47**)



Sol. 259 μM
ThT assay 68% (10 μM)
ThT assay 41% (1 μM)
CC_{50} 149 μM

Sol. 2339 μM
ThT assay 99% (10 μM)
ThT assay 80% (1 μM)
CC_{50} 114 μM

Sol. 2339 μM
ThT assay 99%(10 μM)
ThT assay 75% (1 μM)
CC_{50} 114 μM

Figure 12. Property of diphenyl γ-dihydropyrone type compounds (**47**) and (**53**)

ClogP: c-logarithm partition coefficient, Sol.: water solubility, CC_{50}: cytotoxicity concentration of 50%.

6. BERBERINE AND ITS ANALOGUES

Berberine (**56**) is a benzylisoquinoline alkaloid founds in *Coptis japonica*, an herb whose extracts have long been used in antidiarrheal treatments and eye drops. More recently, berberine (**56**) has demonstrated anti-inflammatory,[54] antibacterial,[55] and neuroprotective effects,[56] and there has been growing interest in its use to treat central nervous system diseases.[57] Structurally, berberine (**56**) incorporates four consecutive rings which are more stable under physiological conditions than curcumin and its analogues; and its methylene acetal group can be regarded as a latent phenolic hydroxy group – the deprotection of which is anticipated to improve its Aβ aggregation inhibitory activity.

The recent focus of our group has been on alkaloids such as galanthamine (**6**),[58] which were recently shown to inhibit Aβ aggregation; and fascaplysin (**54**) and 9-methylfascaplysin (**55**),[59] which recently served as a lead compound substitute for CUR (**1**). Candidate alkaloids are rigid phenyl and/or pyridyl rings and CUR (**1**) has planer structure and sp^2 chain with high flexibility. Since our interest is alkaloid with catechol motif, berberine (**56**) and its derivatives (**57** and **58**) were selected as next targets (Figure 13).

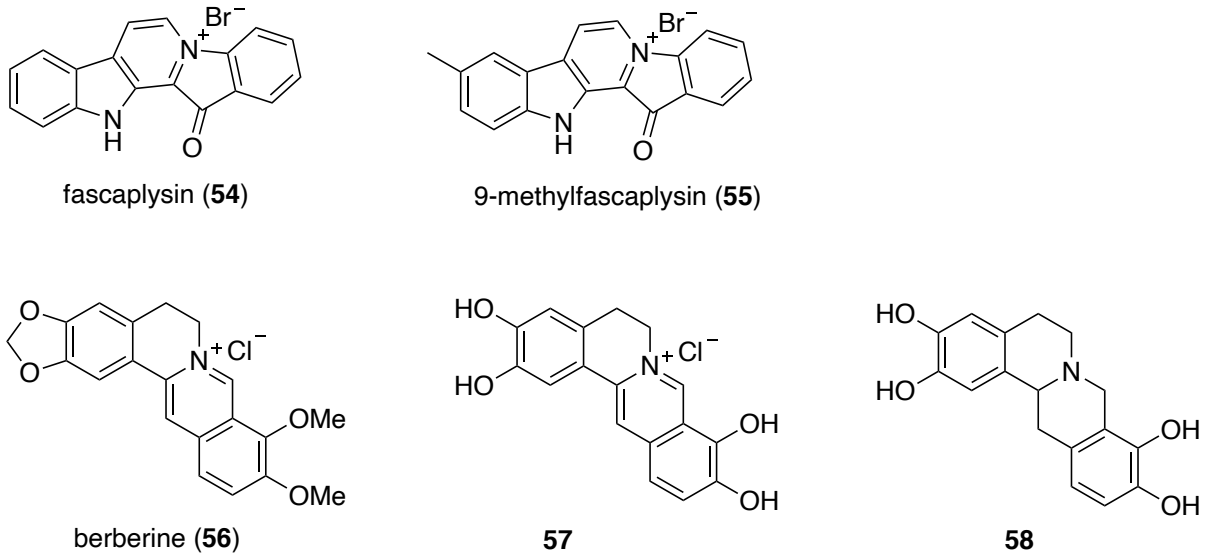
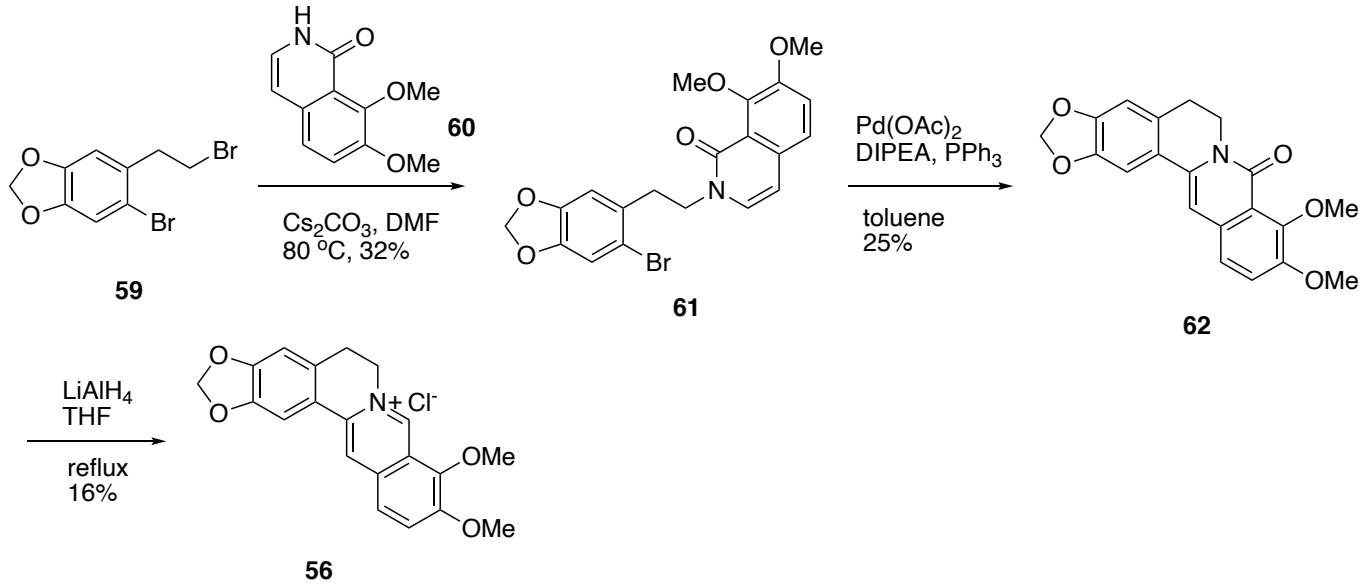


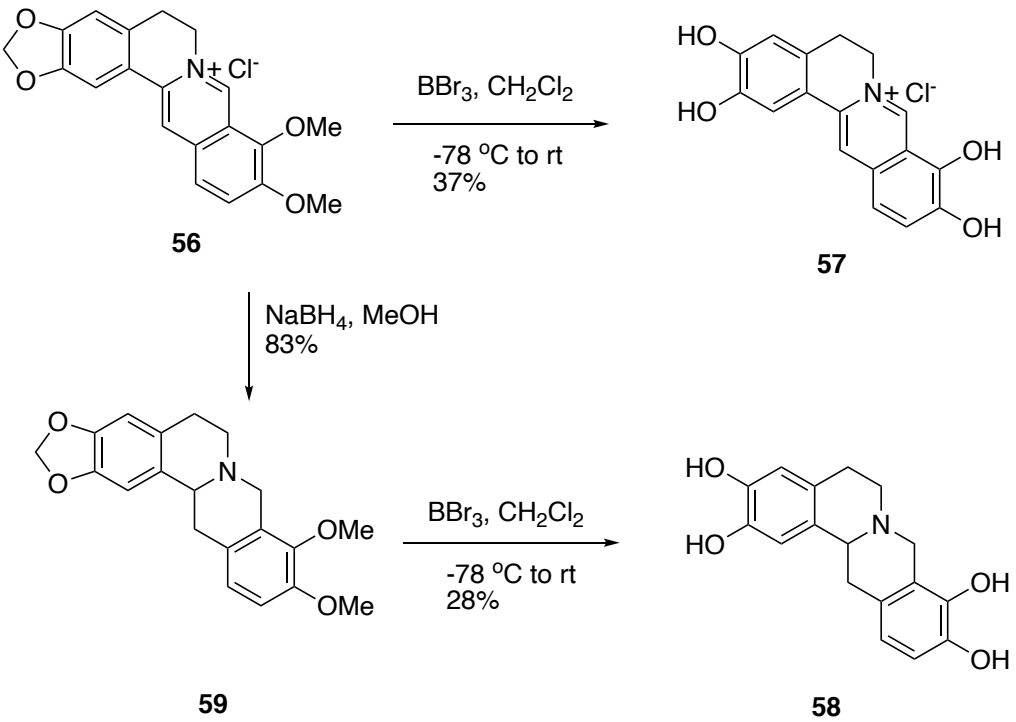
Figure 13. Chemical structures of fascaplysins (**54**) and (**55**) and berberine (**56**) and its analogues (**57**) and (**58**)

We reported total syntheses of berberine (**56**) and selected analogues (**57**) and (**58**)[60] via an intramolecular Heck reaction. *N*-Alkylation of isoquinoline (**59**) and alkyl bromide (**60**) with weak base Cs_2CO_3 in DMF at 80 °C gave the desired product **61** in 32% yield, which underwent the intramolecular Heck reaction using a catalytic amount of $Pd(OAc)_2$ in DIPEA and PPh_3 in toluene to give compound **62** containing the desired berberine framework (**62**) in 25% yield. Reduction of lactam structure of **62** with $LiAlH_4$ in refluxing THF gave berberine (**56**) in 16% yield (Scheme 6).



Scheme 6. Total synthesis of berberine (**56**)

A series of berberine analogues (**57**) bearing varying numbers of phenolic hydroxy groups was prepared from berberine (**56**) by deprotection of the methoxy groups and methylene acetal. Berberine (**56**) was reduced with $NaBH_4$ to obtain quinolizidine (**59**) in 83% yield. Treatment of quinolizidine (**59**) with BBr_3 in CH_2Cl_2 gave tetraol **58** in a yield of 28% (Scheme 7).



Scheme 7. Synthesis of berberine analogues **57** and **58**

Berberine (**56**) was found to exhibit moderate Aβ aggregation inhibitory activity; compound **57** has better water solubility than berberine; and compound **58**, a tertiary amine bearing four hydroxy groups, demonstrated excellent Aβ aggregation inhibitory activity, good water solubility, and low cytotoxicity (Figure 14).

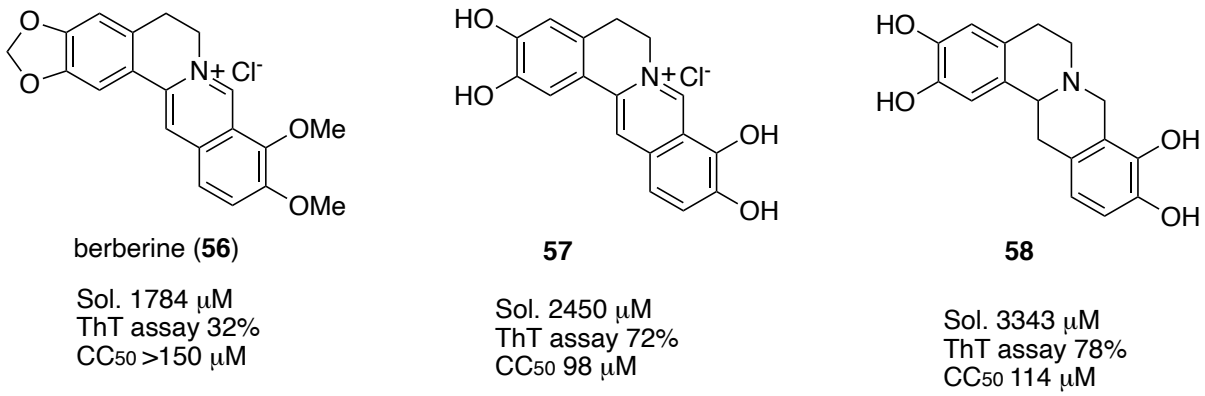


Figure 14. Property of berberine (**56**) and its analogues (**57**) and (**58**)
Sol.: water solubility. CC_{50}: cytotoxicity concentration of 50%.

7. CURCUMIN ANALOGUES AS FLUORESCENCE PROBES

Aβ fibrils are a potential biomarker to diagnose AD and monitor disease progression, and their visualization has been accomplished with a variety of different probes. Congo Red (**64**, CR)[61,62] and ThT (**16**)[63-66] have been used in postmortem histochemical analysis to definitively diagnose AD, and (*trans*, *trans*)-1-bromo-2,5-bis(3-hydroxycarbonyl-4-hydroxy)styrylbenzene (BSB, **65**),[67] and aminophenyl-6-benzothiazole derivatives, *N*-methyl-2-(4-methylaminophenyl)-6-hydroxybenzothiazole (PiB, **66**)[68] have been developed for premortem diagnosis.[69,70] Structurally, BSB (**65**) resembles a 2-carboxy-3-hydroxy derivative incorporating a C8-spacer. Because CUR (**1**) is naturally amyloidophilic and amenable to orally administration, it has been developed as a PET imaging dye or Near-infrared (NIR) dye. ThT (**16**) is the most widely used dye for monitoring amyloid fibrils both *in vitro* and *in vivo*.[71]

Other probes include CRANAD-2 (**67**) and Shiga-Y5 (**68**), which have been used as NIR and ^{19}F-MRI diagnostic imaging probes, respectively. Structurally, they resemble CUR, and their use to detect Aβ fibrils has been studied. As a fluorescent molecular rotor to CUR (**1**),[72] which has five single C-C bonds,[73] Huang et al. demonstrated that (2-phenyl-1,3-benzothiazole)$_2$-iridium(III)-CUR complex (NH-8, **69**) holds promise as a probe for and inhibitor of Aβ aggregation.[74] Compounds **1**, **67** and **70** have also been investigated in the context of prion diseases, whose molecular pathogenesis and neuropathological presentation are similar to AD.[75] Although neither CUR (**1**), CRANAD-2 (**67**), nor CRANAD-5 (**70**) could enhance fluorescence upon binding to amyloid produced by recombinant prion protein, they were found to delay the amyloid formation as measured by the ThT method. The ability to inhibit recombinant PrP

amyloid formation was CUR (**1**) ~ CRANAD-5 (**70**) >CRANAD-2 (**67**). On the other hand, the ability to inhibit the propagation of pathogenic abnormal prion protein in prion-infected cells was CUR (**1**) > CRANAD-2 (**67**) > CRANAD-5 (**70**). This difference in order may be related to differences in actual bioavailability (Figure 15).

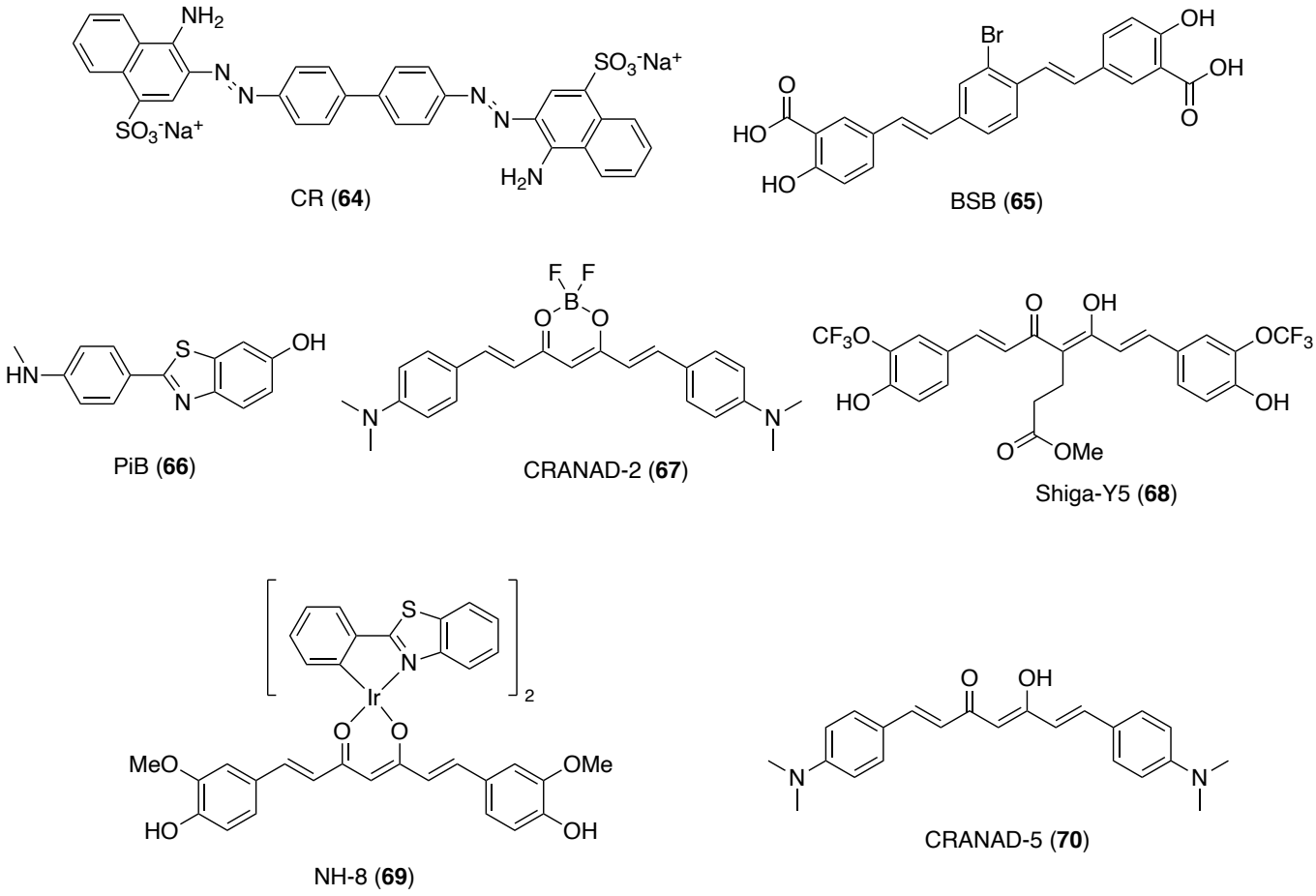


Figure 15. Structures of CR (**64**), CRANAD-2 (**67**), Shiga-Y5 (**68**), BSB (**65**), PiB (**66**), NH-8 (**69**), and CRANAD-5 (**70**)

In the course of our above work, CUR derivative (**71**) was found to fluoresce with an intensity similar to ThT (**16**). Structure-activity studies established the need to protect the phenolic hydroxy groups in order for fluorescence to be observed. In addition, C7-alknenyl chain needs to show this phenomenon from the comparison of downsizing analogues (**72**), (**73**) and (**74**). In addition, Ws-CUR (**75**) was not effective. Development of Ws-CUR (**76**) by our group indicated that asymmetry CUR (**1**) flameworks exploit novel ability. It is anticipated that CUR (**1**) and its analogue (**21**) rarely show aggregation-induced emission (AIE) and can be reversibly showed AIE efficacy depending on the aggregation state of Aβ fibrils. Novel CUR analogues including Me-CUR (**76**) were successfully synthesized and evaluated as new imaging agents for Aβ fibrils. Me-CUR (**76**) showed higher fluorescence than ThT (**16**) as the most extensive amyloid sensor.

Detailed spectroscopic studies indicated that Me-CUR (**76**) has high molecular planarity and thus it could strongly bind with the Aβ fibrils resulting in a large increase in its emission intensity with a large spectral shift. Me-CUR (**76)** is a fluorescent switchable probe to detect Aβ fibrils with high sensitivity (Figure 16, Figure 17).[76]

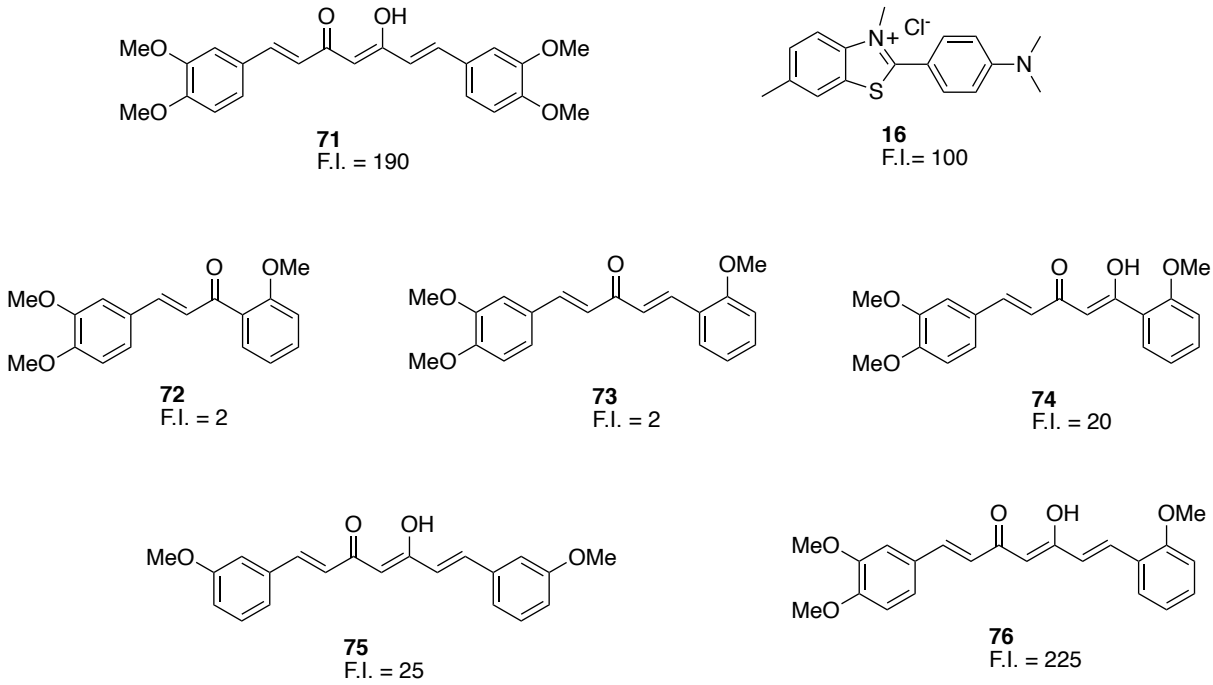


Figure 16. Me-CURs (**71**), (**75**) and (**76**) and their lower molecular weight analogues (**72**), (**73**) and (**74**) F.I.: fluorescence intensity.

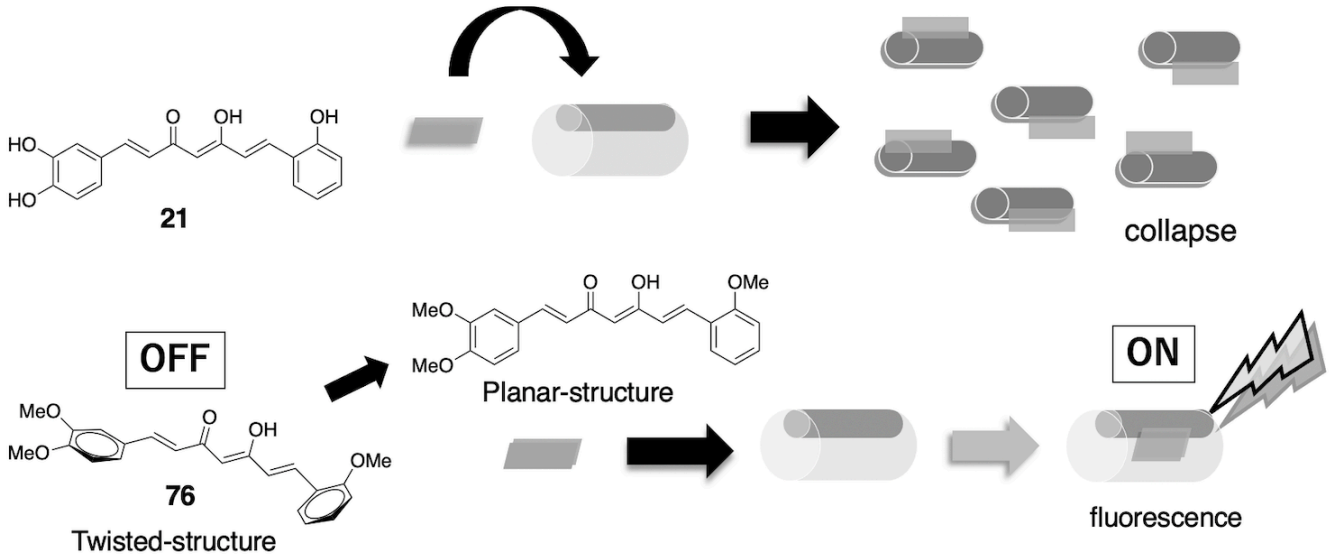


Figure 17. Plausible mechanism of aggregation inhibition and the most stable conformation of Me-CUR (**76**)

8. CONCLUSION

The design, synthesis, and evaluation of catechol-containing compounds for their ability to inhibit and/or detect Aβ aggregation have been reviewed. CUR (**1**) is a well-known natural inhibitor of Aβ aggregation, but its utility is limited by poor bioavailability, a consequence of its extremely low water solubility. We developed catechol derivative (**3**) which is a more water soluble than CUR (**1**) and a more potent inhibitor of Aβ aggregation, by removing the bis-methyl group of CUR (**1**). This use of a catechol moiety to increase the water solubility of Aβ aggregation inhibitors is a general strategy for their design. The positions of the phenolic hydroxy groups on CUR, C5-CUR, diaryl γ-dihydropyrone, and the berberine framework were found to influence their interactions with the Aβ fibrils. This insight resulted in the development of CUR derivative (**21**), C5-CUR derivative (**37**), diaryl γ-dihydropyrone (**47**) and berberine derivative (**57**) as optimized Aβ aggregation inhibitor. We also showed that CUR derivatives could inhibit BACE1. CUR derivative (**76**) that lacks the ability to inhibit Aβ aggregation has potential to detect Aβ fibrils, for diagnostic and disease monitoring purposes. Finally, we showed that CUR (**1**) can inhibit the propagation of pathogenic abnormal prion protein. The derivatives reviewed herein therefore have great potential as treatments and diagnostics for AD.

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REFERENCES

1. M. Urosevic´, L. Nikolic´, I. Gjic´, V. Nikolic´, A. Dinic´, and V. Miljkovic´, *Antibiotics*, 2022, **11**, 135.
2. J.-M. Shieh, Y.-C. Chen, Y.-C. Lin, J.-N. Lin, W.-C. Chen, Y.-Y. Chen, C.-T. Ho, and T.-D. Way, *J. Agric. Food Chem.*, 2013, **61**, 6366.
3. H. Yang, F. Zeng, Y. Luo, C. Zheng, C. Ran, and J. Yang, *Molecules*, 2022, **27**, 38779.
4. L. Lin, Y. Lin, P.-K. Li, J. Fuchs, H. Shibata, Y. Iwabuchi, and J. Lin, *Br. J. Cancer*, 2011, **105**, 212.
5. M. Hashimoto, E. Rockenstein, L. Crews, and E. Masliah, *Neuromol. Med*., 2003, **4**, 21.
6. W. T. Kimberly, J. B. Zheng, S. Y. Guénette, and D. J. Selkoe, *J. Biol. Chem*., 2001, **276**, 40288.
7. Y. Takahara, M. Morishima-Kawashima, Y. Tanimura, G. Dolios, N. Horotani, Y. Horikoshi, F. Kametani, M. Maeda, T. C. Daido, R. Wang, and Y. Ihara, *J. Neurosci.*, 2005, **25**, 436.
8. D. M. Walsh and D. J. Selkoe, *J. Neurochem*., 2007, **101**, 1172.
9. K. Ono, T. Hamaguchi, H. Naiki, and M. Yamada, *BBA Mol. Basis Dis.*, 2006, **1762**, 575.
10. C. Haass and D. J. Selkoe, *Nat. Rev. Mol. Cell Biol*., 2007, **8**, 101.

11. R. Jakob-Roetne and H. Jacobsen, *Angew. Chem. Int. Ed.*, 2009, **48**, 3030.
12. M. Vaz, V. Silva, C. Monteiro, and S. Silverstre, *Clin. Interv. Aging*, 2022, **17**, 797.
13. N. A. Hassan, A. K. Alshamari, A. A. Hassan, M. G. Elharrif, A. M. Alhajri, M. Sattam, and R. R. Khattab, *Molecules*, 2022, **27**, 4839.
14. Z. Q. Zhao, B. Z. Chen, X. P. Zhang, H. Zheng, and X. D. Guo, *Mol. Pharm*., 2021, **18**, 2482.
15. S. Matsuhaba, T. Kishi, and N. Iwata, *PLoS ONE*, 2015, **10**, e0123289.
16. M. Y. Bai, D. B. Lovejoy, G. J. Guillemin, R. Kozak, T. W. Stone, and M. M. Koola, *Complex Psychiatry*, 2021, **7**, 19.
17. J. Eldufani and G. Blaise, *TRCI*, 2019, **5**, 175.
18. F. Yang, G. P. Lim, A. N. Begum, O. J. Ubeda, M. R. Simmons, S. S. Ambegaokar, P. P. Chen, R. Kayed, C. G. Glabe, S. A. Frautschy, and G. M. Cole, *J. Biol. Chem*., 2005, **280**, 5892.
19. J. M. Ringman, S. A. Frautschy, E. Teng, A. N. Begum, J. Bardens, M. Beigi, K. H. Gylys, V. Badmaev, D. D. Heath, L. G. Apostolova, V. Porter, Z. Vanek, G. A. Marshall, G. Hellemann, C. Sugar, D. L. Masterman, T. J. Montine, J. L. Cummings, and G. M. Cole, *Alzheimer's Res. Ther.*, 2012, **4**, 43.
20. M. Ouberai, P. Dumy, S. Chierici, and J. Gaecia, *Bioconjugate Chem.*, 2009, **20**, 2123.
21. S. Dolai, W. Shi, C. Corbo, C. Sun, S. Averick, D. Obeysekera, M. Farid, A. Alonso, P. Banerjee, and K. Raja, *ACS Chem. Neurosci*., 2011, **2**, 694.
22. J. A. Lenhart, X. Ling, R. Gandhi, T. L. Guo, P. M. Gerk, D. H. Brunzell, and S. Zhang, *J. Med. Chem.*, 2010, **53**, 6198.
23. P. Anand, A. B. Kunnumakkan, R. A. Newman, and B. B. Aggarwal, *Mol. Pharm.*, 2007, **4**, 807.
24. R. A. Sharma, W. P. Steward, and A. J. Gescher, *Adv. Exp. Med. Biol*., 2007, **595**, 453.
25. T. M. Kolev, E. A. Velcheva, B. A. Stamboliyska, and M. Spiteller, *Int. J. Quantum Chem*., 2005, **102**, 1069.
26. H. J. J. Pabon, *Recl. Trav. Chim. Pays-Bas*., 1964, **83**, 379.
27. H. LeVine, III, *Protein Sci*., 1993, **2**, 404.
28. K. P. R. Nilsson, A. Aslund, I. Berg, S. Nyström, P. Konradsson, A. Herland, O. Inganäs, F. Stabo-Eeg, M. Lindgren, G. T. Westermark, L. Lannfelt, L. N. G. Nilsson, and P. Hammarström, *ACS Chem. Biol*., 2007, **2**, 553.
29. D. D. Soto-Ortega, B. P. Murphy, J. Gonzalez-Velasquez, K. A. Wilson, F. Xie, Q. Wang, and M. A. Moss, *Bioorg. Med. Chem*., 2011, **19**, 2596.
30. J. Luo, C. Yu, H. Yu, R. Borstnar, S. C. L. Kamerlin, A. Gräslund, J. P. Abrahams, and S. K. T. S. Wärmländer, *ACS Chem. Neurosci*., 2013, **4**, 454.
31. K. Ono, K. Hasegawa, H. Naiki, and M. Yamada, *J. Neurosci. Res*., 2004, **75**, 742.
32. F. Yang, G. P. Lim, A. N. Begum, O. J. Ubeda, M. R. Simmons, S. S. Ambegaokar, P. P. Chen, R.

Kayed, C. G. Glabe, S. A. Frautschy, and G. M. Cole, *J. Biol. Chem*., 2005, **280**, 5892.

33. H. Endo, Y. Nikaido, M. Nakadate, S. Ise, and H. Konno, *Bioorg. Med. Chem Lett*., 2014, **24**, 5621.
34. T. Kakizawa, A. Sanjoh, A. Kobayashi, Y. Hattori, K. Teruya, and K. Akaji, *Bioorg. Med. Chem*., 2011, **19**, 2785.
35. W. Shi, S. Dolai, S. Rizk, A. Hussain, H. Tariq, S. Averick, W. L'Amoreaux, A. E. Idrissi, P. Banerjee, and K. Raja, *Org. Lett*., 2007, **9**, 5461.
36. C. Murray, O. Callaghan, G. Chessari, A. Cleasby, M. Congreve, M. Frederickson, M. Hartshorn, R. McMenamin, S. Patel, and N. Wallis, *J. Med. Chem*., 2007, **50**, 1116.
37. M. Congreve, D. Aharony, J. Albert, O. Callaghan, J. Campbell, R. Carr, G. Chessari, S. Cowan, P. Edwards, M. Frederickson, R. McMenamin, C. Murray, and S. Patel, *J. Med. Chem.*, 2007, **50**, 1124.
38. H. Konno, H. Endo, K. Miyazaki, H. Aoki, A. Sanjoh, K. Kobayashi, Y. Hattori, and K. Akaji, *Bioorg. Med. Chem. Lett*., 2014, **24**, 685.
39. C. W. Murray, O. Callaghan, G. Chessari, A. Cleasby, M. Congreve, M. Frederickson, M. J. Hartshorn, R. McMenamin, S. Petal, and N. Wallis, *J. Med. Chem.*, 2007, **50**, 1116.
40. M. Congreve, D. Aharony, J. Albert, O. Callaghan, J. Campbell, R. A. E. Carr, G. Chessari, S. Cowan, P. D. Edwards, M. Frederickson, R. McMenamin, C. W. Murray, S. Patel, and N. Wallis, *J. Med. Chem.*, 2007, **50**, 1124.
41. H. Hilpert, W. Guba, T. J. Woltering, W. Wostl, E. Pinard, H. Mauser, A. V. Mayweg, M. Rogers-Evans, R. Humm, D. Krummenacher, T. Muser, C. Schnider, H. Jacobsen, L. Ozmen, A. Bergadano, D. W. Banner, R. Hochstrasser, A. Kuglstratter, P. Davis-Pierson, H. Fischer, A. Polara, and R. Narquizian, *J. Med. Chem*., 2013, **56**, 3980.
42. Y.-Z. Xu, S. Yuan, S. Bowers, R. K. Hom, W. Chan, H. L. Sham, Y. L. Zhu, P. Beroza, H. Pan, E. Brecht, N. Yao, J. Lougheed, J. Yan, D. Tam, Z. Ren, L. Ruslim, M. P. Bova, and D. R. Artis, *Bioorg. Med. Chem. Lett*., 2013, **23**, 3075.
43. C. Glaser, *Eur. J. Inorg. Chem*., 1869, **2**, 422.
44. X. Niu, C. Li, J. Li, and X. Jia, *Tetrahedron Lett.*, 2012, **53**, 5559.
45. H. Konno, T. Sato, Y. Saito, I. Sakamoto, and K. Akaji, *Bioorg. Med. Chem. Lett.*, 2015, **25**, 5127.
46. B. List, R. A. Lerner, and C. F. Barbas III, *J. Am. Chem. Soc.*, 2000, **122**, 2395.
47. M. Hotsumi, M. Tajiri, Y. Nikaido, T. Sato, K. Makabe, and H. Konno, *Bioorg. Med. Chem. Lett.*, 2019, **29**, 2157.
48. K. Teruya, S. Iwabuchi, Y. Watanabe, R. Tsuchida, M. Watanabe-Matsui, H. Konno, and K. Doh-ura, *Biochim. Biophys. Acta, Gen. Subj.*, 2022, **1866**, 130094.
49. F. Kiuchi, Y. Goto, N. Sugimoto, N. Akao, K. Kondo, and Y. Tsuda, *Chem. Pharm. Bull.*, 1993, **41**, 1640.

50. A. Simon, D. P. Allais, J. L. Duroux, and J. P. Basly, *Cancer Lett.*, 1998, **129**, 111.
51. R. Adhikary, C. A. Barnes, R. L. Trampel, and S. J. Wallace, *J. Phys. Chem. B*, 2011, **115**, 10707.
52. R. A. Khera, R. Ahmad, I. Ullah, O.-U.-R. Abid, O. Fatunsin, M. Sher, A. Villinger, and P. Langer, *Helv. Chim. Acta*, 2010, **93**, 1705.
53. M. Hotsumi, M. Tajiri, K. Makabe, and H. Konno, *Bioorg. Chem.*, 2020, **104**, 104302.
54. C.-L. Kuo, C.-W. Chi, and T.-Y. Liu, *Cancer Lett.*, 2004, **203**, 127.
55. F. R. Stermitz, P. Lorenz, J. N. Tawara, L. A. Zenewicz, and K. Lewis, *Proc. Natl. Acad. Sci. USA*, 2000, **97**, 1433.
56. M. L. Aski, M. E. Rezvani, M. Khaksari, Z. Hafizi, Z. Pirmoradi, S. Niknazar, and F. Z. Mehrjerdi, *Iran. J. Basic Med. Sci.*, 2018, **21**, 53.
57. J. Zhang, J.-Q. Yang, B.-C. He, Q.-X. Zhou, H.-R. Yu, Y. Tang, and B.-Z. Liu, *Saudi Med. J.*, 2009, **30**, 760.
58. B. Matharu, G. Gibson, R. Parsons, T. N. Huckerby, S. A. Moore, L. J. Cooper, R. Millichamp, D. Allsop, and B. Austen, *J. Neurol. Sci.*, 2009, **280**, 49.
59. Q. Sun, F. Liu, J. Sang, M. Lin, J. Ma, X. Xiao, S. Yan, C. B. Naman, N. Wang, S. He, X. Yan, W. Cui, and H. Liang, *Mar. Drugs*, 2019, **17**, 121.
60. M. Tajiri, R. Yamada, M. Hotsumi, K. Makabe, and H. Konno, *Eur. J. Med. Chem.*, 2021, **215**, 113289.
61. K. Blennow, M. J. de Leon, and H. Zetterberg, *Lancet*, 2006, **368**, 387.
62. S. D. Selkoe, *Physiol. Rev.*, 2001, **81**, 741.
63. A. C. Mathis, J. B. Bacskai, T. S. Kajdasz, E. M. McLellan, P. M. Froach, T. B. Hyman, P. D. Holt, Y. Wang, G. F. Huang, L. M. Debnath, and E. W. Klunk, *Bioorg. Med. Chem. Lett.*, 2002, **12**, 295.
64. A. Nordberg, *Lancet Neurol.*, 2004, **3**, 519.
65. P. Divry, *J. Belge Neurol.*, 1927, **27**, 643.
66. G. Kelényi, *Acta Neuropathol.*, 1967, **7**, 336.
67. D. M. Skovronsky, B. Zhang, M. P. Kung, H. F. Kung, J. Q. Trojanowski, and V. M. Lee, *Proc. Natl. Acad. Sci. USA*, 2000, **97**, 7609.
68. A. L. Boxer, G. D. Rabinovici, V. Kepe, J. Goldman, A. J. Furst, S.-C. Huang, S. L. Baker, J. P. O'Neil, H. Chui, M. D. Geschwind, G. W. Small, J. R. Barrio, W. Jagust, and B. L. Miller, *Neurology*, 2007, **69**, 283.
69. S. Furumoto, N. Okamura, R. Iwata, K. Yanai, H. Arai, and Y. Kudo, *Curr. Top. Med. Chem.*, 2007, **7**, 1773.
70. K. Teruya and K. Doh-ura, *Curr. Top. Med. Chem.*, 2013, **13**, 2522.
71. S. H. Hudson, H. Ecroyd, W. T. Kee, and A. J. Carver, *FEBS J.*, 2009, **276**, 5960.
72. J. Bisson, J. B. McAlpine, J. B. Friesen, S.-N. Chen, J. Graham, and G. F. Pauli, *J. Med. Chem.*, 2016,

59, 1671.

73. I. Toyama, D. Yanagisawa, H. Taguchi, T. Kato, K. Hirao, N. Shirai, T. Sogabe, N. F. Ibrahim, T. Inubushi, and S. Morikawa, *Aging Res. Rev.*, 2016, **30**, 85.
74. Y. Huang, H. Na, L. Sun, K. Terpstra, K. Gui, Z. Yu, and L. M. Marica, *Chem. Rxiv.*, 2021, DOI.10.26434/chemrxiv.13606034.v1.
75. S. B. Prusiner, M. P. McKinley, K. A. Bowman, D. C. Bolton, P. E. Bendheim, D. F. Groth, and G. G. Glenner, *Cell*, 1983, **35**, 349.
76. T. Sato, M. Hotsumi, K. Makabe, and H. Konno, *Bioorg. Med. Chem. Lett.*, 2018, **28**, 3520.

Hiroyuki Konno was born in Miyagi, Japan. He completed his doctoral work in 1999 at the Tohoku University in the supervision of Dr. Kunio Ogasawara. After working as a postdoctoral fellow at the University of Pennsylvania, USA, he worked as an Assistant Professor at University of Tokushima, Kyoto University, and Kyoto Prefectural University of Medicine. He joined Yamagata University as an Associate Professor in 2009 and was promoted to Professor in 2018. His research interests focus on natural product synthesis, peptide chemistry, and the creation of enzyme inhibitors.

Kenta Teruya was born in Hyogo, Japan. He completed his doctoral work in 2002 at Osaka University under the supervision of Dr. Saburo Aimoto. After working as a postdoctoral fellow at the National Institutes of Health, National Cancer Institute, USA, he worked as an Assistant Professor at Tohoku University in 2003. He joined Kyoto Prefectural University of Medicine in 2010 and then Tohoku University again in 2015 as an Associate Professor. His research interests are prion biology, peptide and protein chemistry, and the discovery of drugs to treat prion diseases.